

Reviewer1:

This manuscript reports interesting results from experiments designed to investigate organic matter degradation dynamics in sediments from the Arabian Sea. The authors have found an interesting de-coupling of degradability from conventional measures of organic matter composition/quality, and this merits publication. I feel however that some aspects of the manuscript need to be revised before publication, to ensure that the mechanisms behind the observed trends are fully explored and discussed.

Authors' response:

The authors thank the reviewer for the general positive response on the manuscript and the thorough review of it.

Main Comments

Reviewer1:

Introduction. The introduction provides a good review of the appropriate literature, but does not really identify a research gap, or any research questions. It would be helpful if an explanation was added as to what motivated the study.

Authors' response:

A research question/study motivation will be added to the revised manuscript. Emphasis will be given to examination of drivers and constrains of OM degradation in marine sediments. In addition, the fact that despite several existing studies on biochemical quality of OM in Arabian Sea sediments (e.g. Cowie and Levin 2009, Vandewiele et al. 2009, Woulds and Cowie, 2009) or experimental approach investigating microbial degradation of OM (Moodley et al. 2011), the combination of these studies is lacking.

Reviewer1:

The last section of the introduction tells the reader what work was done, but could also summarise the main finding.

Authors' response:

We feel that the introduction is not the appropriate place for a summary of results (this is already done in the abstract). The introduction should provide an overview of study subject and previous work related to it. Study aims/motivations are often given at the end of the introduction.

Reviewer1:

Section 3.2. I feel that the main issue here is the question of the extent to which the fact that all incubations were conducted under oxic conditions has affected the results. Presumably for oxic degradation to have occurred in sediments from the OMZ a new microbial community had to develop first. I realise that you have already acknowledged this point; however I feel that it warrants further discussion in light of the literature. For example, what do we know about whether oxic microbes are even present in OMZ sediments, and how long would it take them to develop into a fully functional community? How are the duration and conditions of storage of samples before incubation likely to have affected the microbial communities in samples from both within and outside the OMZ?

Authors' response:

Throughout the manuscript we report the microbial respiration rates as the *potential* mineralisation rates and nowhere we indicate that these are the rates found in situ. As the reviewer also points out we have already outlined the main concerns regarding the

oxic incubations of the OMZ sediments. However, to further clarify our approach we refer to relatively recent papers by Fenchel and Finlay (2004), and De Wit and Bouvier (2006) that elaborate on the statement of Baas Becking “Everything is everywhere, but, the environment selects”, implying that all bacteria are found everywhere and environment, here oxic, will activate the “right” community. It is true that this may take some time. But our remineralisation experiment ran for nearly 3 weeks and this should provide enough time for a community to function accordingly.

Reviewer1:

I feel that the paper currently does not really come to a firm conclusion; therefore some might say that it is not clear what the central finding or idea is. The lack of correlation between biochemical quality and microbial degradability is intriguing and worthy of publication, however I feel that it requires considerable further discussion. It would be best if that discussion yielded a suggestion from you as to which of the mechanisms you discussed are actually controlling degradability. This discussion should include acknowledgement of the fact that previous degradation experiments have found that biochemical quality or freshness was linked to degradability or half life.

Authors' response:

Reviewers' comment will be addressed in the revised version and a firmer conclusion will be drawn. In the submitted version we discussed possible control mechanisms in section “3.3 OM bioavailability versus biochemical OM quality” where at the end we also suggest a link with macrofaunal processing of OM, organic matter being more available to microbes if processed by macrofauna. We will also add to the discussion the new study of Hunter et al. (2012) as suggested by reviewer#2.

Reviewer1:

The sections on macrofaunal populations and bioturbation (page 12 onwards) do not currently seem to serve a purpose. This is particularly true for the bioturbation section, which seems to report your mixing data without relating this back to the central question of how it might have affected degradability.

Authors' response:

As we have no data for macrofaunal abundance for most stations and we suggest a link between OM bioavailability and macrofaunal processing, we used sediment mixing/bioturbation to indicate “impact” of macrofauna on sediment processing. Although bioturbation is present throughout the study transect, it is minimal in the core of the OMZ and higher at the better-ventilated sites. Thus, we believe that the bioturbation data are central for the paper. In the revised version we will integrate this section better with the discussion of OM bioavailability.

Other Comments:

Reviewer1:

Introduction paragraph 1. It would be worth mentioning here that a significant number of studies have found oxygen concentration not to be a primary control on OM degradation/preservation. Canfield (1994) produced the best resolution of the two sides of the argument, with his emphasis on the importance of oxygen exposure time. This is particularly worth mentioning because at the start of section 3.3 you also appear to suggest that straightforward oxygen availability is not necessarily the main factor driving OM accumulation in OMZs.

Authors' response:

The Canfield (1994) paper will be added to the resubmission

Reviewer1:

Methods. Make sure that OMZ is defined the first time it is used.

Authors' response:

OMZ will be defined when used for the first time.

Reviewer1:

I suggest additional proof reading, as there are several locations where words such as 'a' and 'the' have been missed out (e.g. page 5 line 10 'we performed series of sediment incubations...').

Authors' response

Proof reading will be done before resubmission.

Reviewer1:

Section 2.1. I feel that the discussion of whether microbial communities will have changed due to experiments being conducted under oxic conditions should also acknowledge that communities will have altered during sample storage for 2 months.

Authors' response

The active community may have changed during sample storage. Still, this would be the case for all samples and hence should not have altered our findings substantially. Also see previous authors' response (above) regarding the bacterial communities in the experimental slurries.

Reviewer1:

It would be helpful to know what conditions the samples were stored in (oxic or anoxic?).

Authors' response

Samples were stored in plastic bags at 4°C, which were sealed immediately after slicing. No oxygen content was measured during storage but anoxic conditions may be anticipated due to lack of aeration. Additional information will be added about the experimental methods in the final resubmission as also suggested by reviewer#2.

Reviewer1:

Page 5 last line: Please correct units in the phrase 'per wet sediment'.

Authors' response

Corrected. Units "ml" added

Reviewer1:

Methods: I feel as though I need a little more detail on how the sediment incubations were carried out, e.g. how many replicate incubations per site, what volume of sediment was used, and how were oxic conditions maintained?

Authors' response

More detail will be added to resubmitted manuscript. Here in short:

Two replicates were used. The incubations were carried out in 80 ml bottles. 10 ml of sediment was inserted into each bottle that were subsequently filled with well-aerated 0.2 µm filtered seawater (low-nutrient deep Atlantic water). Total water volume, as well as accurate conversion of wet and dry weight sediment, was obtained by direct weighing. Throughout the experiment the bottles were periodically shaken to mix the

slurry. At the end of the incubations the oxygen content was measured with an oxygen optode (Presens, Germany). The oxygen content in each bottle at the end of the incubation was always $>20 \mu\text{M}$.

Reviewer1:

Section 2.2. I suggest including a little more detail here on THAA analysis, such as the fact that you produced acid hydrolysates from sediment samples and analysed them by HPLC. The same applies to PLFA analysis.

Authors' response

THAA and PLFA methods will be explained in more detail in the resubmission.

Reviewer1:

Methods: I would suggest that some phaeopigments seem to have half lives of thousands of years (see reference in Woulds and Cowie 2009), which sheds doubt on your statement that downcore penetration of phaeopigments could only occur due to faunal mixing.

Authors' response

We decided not to report half-lives of pigments in this study (although the reviewer is correct in saying that if calculated they would be on the order of 1000s of years). Due to bioturbation at all of our study sites, as shown by the ^{210}Pb and pigment data, it is not possible to discriminate between pigment degradation and downward mixing of pigments by animals and consequently calculations of half-lives would be strongly biased.

Reviewer1:

Please state how pigment inventories were calculated.

Authors' response

The pigment inventories were calculated as an integrated sum of pigments in top 10 cm of sediment.

Reviewer1:

C accumulation rates. Please state how %C_{org} values were measured. I would also like to see further justification of the use of C_{org} data from the top three cm for calculating burial rates. I can see your point within the OMZ (although readers who have not worked in OMZs might not be able to take your point as read), but I remain sceptical that your approach is valid below the OMZ at oxic sites. I suggest that you describe the maximum downcore decrease in %C_{org} that you saw at an oxic site, and demonstrate how much difference it would actually make to your estimations of C burial if you used %C_{org} data from say 20 cm instead of 0-3 cm.

Authors' response

The %C_{org} was measured on ground freeze-dried sediments (from the slurry sample; top 3 cm) applying conventional methods. Organic carbon and nitrogen contents were measured using an elemental analyzer following acidification to remove any carbonate from sediment (Nieuwenhuize et al., 1994).

As the burial rates are based on the average top 3 cm value, and not on top 1 cm value for example, the typical decrease in C_{org} content, which may be anticipated at the oxic sites is already averaged out in our stations. In fact, when using the C_{org} content of sediment slice 18-20 cm to calculate burial rates at most sites a slight increase is observed, although the pattern remains the same. The only station where burial rates

are somewhat lower, if the burial calculation is based on a deeper horizon, was OMZ station PA-2. In addition, a point to note is that the burial rate calculation is not only based on C_{org} content but also on the porosity, which is lower in the deeper sediment samples. As the trend in the burial calculations remains the same if using the top 3 cm or deeper horizon, and as an increase rather than decrease is seen in the burial rates (in contrast to reviewer expectations), and as the typical decline in the C_{org} profiles takes place within the top 3 cm, we believe that our burial estimates are valid in their current form.

Reviewer1:

Results. You could try referring to your stations by their depths, rather than by numbers (which although logical are still arbitrary). This might help your reader keep track of which site is which (although this is only a suggestion, as it is simply the system I am used to using).

Authors' response

Station names will be changed to water depths in the resubmission.

Reviewer1:

Page 9, paragraph 1: I think the relationships referred to as correlations here are actually regressions, as R^2 values are stated. Please correct/clarify this. Please also state the p values for these relationships to show that they are statistically significant.

Authors' response:

Correct term is regression as the reviewer points out. This will be corrected in the resubmission. The p -values are, O_2 and DI ($p < 0.001$; or exact 0.000006); O_2 and intact/total pigments ($p = 0.02$) and DI and O_2 ($p = 0.009$). P -values will be added to the resubmission.

Reviewer1:

Figure 3. Please add an explanation of what the closed and open circles in panel G mean.

Authors' response

Closed (bacterial biomass) and open circle (mineralisation rate) are explained in the figure already (next to the axis). Additional explanation will be given in figure caption.

Reviewer1:

Section 3.2. The two most oxygenated sites seem to show bacterial biomass considerably lower than any other values. I feel this should be acknowledged in this section (even though it probably doesn't make a difference to your data interpretation).

Authors' response

We agree that the bacterial biomass at station 9 ($127 \text{ mmol C m}^{-2}$), the second most oxygenated site, is considerably below the average. However at the deepest site the bacterial biomass is $214 \text{ mmol C m}^{-2}$, which matches well with the average $248 \pm 67 \text{ mmol C m}^{-2}$. The minimum biomass at site 9 is acknowledged in the revisions of the manuscript.

Reviewer1:

Page 11 line 25: Please further explain what you mean by '...macrofauna may provide catalyzers for microbial degradation...'

Authors' response

The sentence on this current form is confusing as the reviewer points out. In the resubmission it will be modified to “This way macrofauna may provide catalyzers for microbial degradation, thus simultaneously aiding in the breakdown of macromolecular compounds.” So “in addition” will be replaced by “this way”.

Reviewer1:

Page 12 line 25: ‘inferred’ rather than ‘induced’.

Authors’ response

This will be changed in resubmission.